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SEROLOGICAL AND PRODUCTION RESPONSES TO VACCINATION AGAINST RESPIRATORY PATHOGENS AT BRANDING AND WEANING IN BEEF CALVES

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Serological and Production Responses to Vaccination against Respiratory Pathogens at Branding and Weaning in Beef Calves

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Abstract:
The objectives of this project were to determine if vaccination of young calves against various respiratory pathogens provides additional protection to calves post-weaning after being given a booster vaccination at weaning and to determine the nature of the serological response to vaccines in young calves which are vaccinated at branding. 290 calves from the Western Beef Development Center were followed. Calfhood vaccination prior to pasture turnout with a Mannheimia hemolytica Leukotoxin/Hemophilus somnus vaccine seemed to prime the immune response of these calves so that at weaning they had a higher immune response when boostered with the same vaccination.

Executive Summary:

Pre-vaccination calf programs have been promoted by a number of pharmaceutical companies in the past few years. These programs usually rely on a spring calfhood vaccination which is meant to "prime" the immune system of the calf so that vaccinations at weaning in the feedlot would have a better and faster response due to immunological memory. Another potential benefit might be to mitigate the occurrence of enzootic pneumonia or "summer pneumonia" of young calves on pasture as their immunity wains. However, there are relatively few field trials that have documented this immune response and whether or not it is protective.

The objectives of this trial were to:

1) To determine if vaccination of young calves at branding against various respiratory pathogens provides protection against respiratory disease over the summer grazing season.

(2) To determine if vaccination of young calves at branding against various respiratory pathogens provides additional protection to calves post-weaning after being given a booster vaccination at weaning.

(3) To determine the nature of the serological response to vaccines in young calves which are vaccinated at branding.

(4)To determine the effects of vaccination on sub-clinical lung lesions at slaughter and the effect of these lesions on performance and carcass quality.

(5) To examine the effects of modified live vs. killed viral vaccines in well vaccinated calves.

The calf crop from the Western Beef development centre were the animals used for this trial. Just prior to pasture turnout, at "branding time" the calves were sorted into four distinct treatment groups which consisted of different vaccination programs. All calves received clostridial vaccinations and routine dehorning and

surgical castration occurred at this time as well. The four treatment groups are summarized in the following table.

Table 1: Vaccinations given to calves in each treatment group

TIMING	Spring Branding		Fall Weaning		Post-Weaning	
Date	June 12 & 13		Oct 1 & 2		Oct 23 & 24	
VACCINE	SS-PH	SV4	SS-PH	SV4	SS-PH	SV4
Treatment A	X		X	X		
Treatment B		X		X		
Treatment C	X	X	X	X		
Treatment D			X	X		X

Vaccines:

SS-PH: (Somnustar-PH, Novartis Animal Health Canada Inc.) A killed vaccine which contains antigens for Hemophilus somnus and Pasteurella hemolytica

SV4: (StarVac 4 Plus, Novartis Animal Health Canada Inc.) A modified live vaccine which contains antigens for bovine viral diarrhea virus (BVD) type I and II, Infectious bovine rhinotracheitis virus (IBR) , bovine respiratory syncytial virus (BRSV) and parainfluenza 3 (PI3) virus.

The calves were followed throughout the summer and after weaning were backgrounded at the Termeunde farm. After backgrounding, the calves were transferred to the Poundmaker feedlot for finishing. Treatment records were maintained on calves throughout the trial. Blood samples were collected at branding, weaning and post weaning. In calves from group D a fourth blood sample was collected after their post-weaning booster vaccination (6 weeks post weaning).

Unfortunately, Canada announced it's first indigenous case of bovine spongiform encephalopathy on May 20, 2003. This event obviously had major implications on the beef industry in general and as a result impacted this study. The cattle on this trial were still on feed at the feedlot and as a result of the effects of BSE on the Canadian cattle market, the cattle were not finished and marketed in a normal scenario. This meant that the carcass data and lung lesion scores were no longer useful as the animals had remained on feed for a much longer period of time than was normal. As well, market conditions necessitated marketing these animals at very irregular intervals which meant that the logistics of collecting this data became very difficult. As a result, we were unable to complete the objectives related to carcass data and lung lesion scores.

There were relatively few treatments for respiratory disease among this group of cattle. All of the treatments occurred during the feeding phase of the trial and

only 9.7% of the calves on average were treated for bovine respiratory disease. These treatments were distributed relatively evenly across all vaccination programs. Only 3 calves died during the trial including a broken leg, and a bloat case. One calf died as a result of chronic bovine respiratory disease.

Calves in treatment group A tended to have a higher post-weaning immune response to two important respiratory pathogens. The titers to Hemophilus somnus and Mannheimia hemolytica Leukotoxin were both significantly higher. Calves in treatment group C also tended to have slightly higher post-weaning immune responses although these were not statistically higher.
Hemophilus somnus causes a variety of diseases and is seen primarily in feedlot cattle. Hemophilus somnus is associated with approximately 1/3 of all feedlot cattle mortalities. Mannheimia hemolytica is a bacterial agent that was formerly called Pasteurella hemolytica. It is the bacteria that is the primary cause of fibrinous pneumonia in feedlot cattle or "shipping fever".

These results would suggest that calves which receive Somnustar PH (Novartis Animal Health Canada Inc.) as a calfhood vaccination have their immune systems primed to these two agents of respiratory disease. As a result, when they receive a second dose at weaning time, their immune response is higher than calves that did not receive the vaccine at spring turnout. This might suggest that these calves would be better protected against disease caused by these bacteria during the early feeding period. This "memory" response in these calves was similar to the memory response we see in calves which received the vaccine at weaning and then again 3 weeks later (Group D calves).

A second trial was performed on calves in Group A and C. 45 calves from these groups were randomly selected later in the feeding period (147 days or almost 5 months after weaning). All of these calves had already received two doses of Somnustar PH and at least one dose of StarVac 4 Plus (Novartis Animal Health Canada Inc.). These calves were divided into three groups, 5 calves received a placebo made of sterile diluent, 20 calves received another dose of StarVac 4 Plus (the modified live vaccine), and 20 calves received a dose of ViraShield 5 (a killed respiratory vaccine containing IBR, PI3, BVD type 1 and 2 and BRSV). Blood samples were collected at the time of vaccination and 3 weeks later. There was no difference in titers prior to this vaccination and the calves had low to moderate titers to BVD 5 months after weaning. Calves which received the killed vaccine had a much higher immunological response to IBR, BVD type 1 and 2 and to BRSV than calves which received another dose of modified live vaccine.

Information of benefit to producers

This trial demonstrates the benefits of utilizing a Mannheimia hemolytica leukotoxin/Hemophilus somnus vaccine in a calfhood vaccination program. Although there was no enzootic pneumonia present in this group to evaluate the

effects on young calves, it would appear that giving Somnustar PH (Novartis Animal Health Canada Inc.) does prime the immune system so that these calves respond with memory to a second vaccination at weaning. This has implications in terms of providing value to the feedlot in terms of prevaccination programs and also has a benefit to producers who are retaining ownership of calves.

It also demonstrated that in calves who have already received a modified live viral vaccine that a booster vaccination with a killed vaccine produces a significantly higher titer response than a booster vaccination with a modified live vaccine. This has significant implications for feedlots who are purchasing pre-vaccinated calves. It also has an implication for heifer replacement vaccination programs and perhaps even adult cow vaccination programs. These findings need to be investigated in greater detail.

Technical Report

Introduction

Respiratory disease is a major cause of morbidity and mortality in cattle. Summer pneumonia in calves or "Enzootic pneumonia" is an important disease in Saskatchewan community pastures. This disease often occurs at two to six months of age when maternal-derived immunity wanes. Ideally, a method to improve the immunity during this risk period is needed to prevent this disease. Although maternal immunity may interfere with a young calf's response to vaccination, recent studies suggest that immunization with some vaccines may prime the immune cells so that when the calf is exposed to the disease or is re-vaccinated there is a memory response of the immune system.

Respiratory disease in feedlots accounts for 75% of illnesses and 45% to 55% of the population mortality. Controlling respiratory disease by vaccination at arrival has proven to be difficult at best. The vast majority of cases occur in the first few weeks after arrival which means that traditional vaccination programs do not have enough time to mount an adequate immune response. Lung lesions at harvest have been correlated to performance and carcass quality. The control of these diseases through effective vaccination protocols would provide an economic benefit to Saskatchewan cattle producers. Controlling these diseases through vaccination would also decrease the use of antibiotics in these populations and decrease injection site lesions. Antibiotic usage in food animals and the development of antimicrobial resistance is an important public health concern.

There have been reports that cattle vaccinated with Modified Live Viral Vaccines (MLV) have an erratic and sub-optimal antibody response to subsequent boosters with a Modified Live Viral Vaccine. Vaccine induced interference is thought to be do a combination of cell mediated immunity stimulated by previous vaccination and circulating antibody limiting the replication of vaccine virus and immune stimulation. Inactivated vaccines appear to stimulate an effective antibody response following an appropriate vaccination protocol with either a MLV or an inactivated vaccine. Inactivated vaccines contain a high antigenic mass and are not thought to be affected by a cell-mediated response. An inactivated vaccine may be a more effective annual booster providing a more consistent response resulting in higher antibody levels than a MLV. There are numerous references in the literature regarding boosting MLV with an inactivated vaccine, but it is difficult to find published data substantiating the protocol.

The objectives of this project were to:

(1) To determine if vaccination of young calves at branding against various respiratory pathogens provides protection against respiratory disease over the summer grazing season.

(2) To determine if vaccination of young calves at branding against various respiratory pathogens provides additional protection to calves post-weaning after being given a booster vaccination at weaning.

(3) To determine the nature of the serological response to vaccines in young calves which are vaccinated at branding.

(4)To determine the effects of vaccination on sub-clinical lung lesions at slaughter and the effect of these lesions on performance and carcass quality.

(5) To examine the effects of modified live vs. killed viral vaccines in well vaccinated calves.

Materials and Methods:

The cattle were crossbred Angus calves 9 – 11 months old resident at the Western Beef Development Centre, Trumunde Farm, Lanigan, Saskatchewan. The calves were born on the farm, spent the summer on pasture at the farm or at Pathlow Pasture in northeast Saskatchewan. On June 12th and 13th, 2002 the calf crop of the Western Beef Development Centre were processed at branding time (ie: prior to turnout onto pasture). All calves received a multivalent clostridial bacterin subcutaneously. Bull calves were surgically castrated and horned calves were dehorned. Two hundred and ninety calves were randomized

using a random number generator into one of four treatment groups. Blood samples were collected from forty randomly selected calves in each treatment group. Vaccinations were given subcutaneously to every calf in each treatment group as follows:

Group A: Somnustar PH (Novartis, Animal Health Canada, Inc.)
Group B: StarVac 4Plus (Novartis, Animal Health Canada, Inc.)
Group C: StarVac 4Plus + Somnustar PH (Novartis, Animal Health Canada, Inc.)
Group D: Placebo

These calves were then turned out to pasture with their dams. Treatment records were maintained for any calves that were sick and treated by farm staff. Farm staff were blinded to which vaccines the calves received.

On October 1 and 2, the calves were weaned and processed. All calves received a subcutaneous multivalent clostridial bacterin. Blood samples were collected again from the 120 calves that had been sampled at branding. Calves in each treatment group were given all vaccinations subcutaneously as follows:

Group A: Somnustar PH + StarVac4Plus (Novartis, Animal Health Canada, Inc.)
Group B: StarVac 4Plus (Novartis, Animal Health Canada, Inc.)
Group C: StarVac4Plus+Somnustar PH (Novartis, Animal Health Canada, Inc.)
Group D: StarVac 4Plus (Novartis, Animal Health Canada, Inc.)

Vaccination sites were palpated approximately 2 days after weaning.
On October 23 and 24th, blood samples were collected again from the same randomly selected calves in each treatment group. Calves in Group D were given a booster vaccination of StarVac 4Plus (Novartis, Animal Health Canada, Inc.). Table 1 summarizes the vaccinations given to each treatment group.

On November 13th, blood samples were collected for the final time from randomly selected calves in Group D.

These calves were backgrounded at the Western Beef Development Centre and were then transferred to Poundmaker Feedlots for finishing. Treatment records were maintained throughout the life of each calf. The calves were commingled for the duration of the trial. The calves were housed in a typical backgrounding lot, open air, dirt floor pen, with a porosity fence for a windbreak, and a centre facing alley with a concrete feed bunk.

Unfortunately, Canada announced it's first indigenous case of bovine spongiform encephalopathy on May 20, 2003 This event obviously had major implications on the beef industry in general and as a result impacted this study. The cattle on this trial were still on feed at the feedlot and as a result of the effects of BSE on the Canadian cattle market, the cattle were not finished and marketed in a normal scenario. This meant that the carcass data was no longer useful as the

animals had remained on feed for a much longer period of time than was normal. As well, market conditions necessitated marketing these animals at very irregular intervals which meant that the logistics of collecting this data became very difficult. As a result, we were unable to complete the objectives related to carcass data and lung lesion scores.

To accomplish objective 5, 45 calves were randomly selected from Treatment Groups A or C. This occurred later in the feeding period at 147 days following the weaning vaccination. Both of these groups had previously received at least one dose of a modified live viral vaccine (Star Vac 4 Plus) (Novartis, Animal Health Canada, Inc.)at weaning, and all had received Somnustar PH (Novartis, Animal Health Canada, Inc.) in the spring as young calves. These calves were randomized into 3 groups by a computer generated random number. Five calves received a placebo consisting of a sterile diluent, 20 calves received a killed vaccine containing IBR, PI3 virus, BVD type 1 and BVD type 2, and BRSV (Virashield 5) (Novartis, Animal Health Canada, Inc.), while the other 20 calves received a modified live vaccine which contained the same viral components (StarVac 4 Plus) (Novartis, Animal Health Canada, Inc.). Calves were bled at the time of vaccination and 21 days post vaccination (Day 168 post weaning) and vaccination titres were compared.

Statistical Analysis:

Chi-square analysis was used to examine differences in morbidity and mortality in treatment groups. Kruskal Wallis one-way ANOVA was used to examine for statistical differences between treatment groups with regard to titre values. A simple one-way ANOVA was used to evaluate treatment differences in weight gain.

Results:

Weaning Weight and Health Data:

There were no differences in spring calf weights or in fall weaning weights between any of the treatment groups. Spring calf weights averaged 197.4 lbs (SD = 36.0) and fall weaing weights averaged 490.0 lbs (SD=59.2) Morbidity due to bovine respiratory disease (BRD) was low for this group of calves as only 9.6% of them were treated during the trial. All of these treatments occurred during the feedlot phase of the trial. Morbidity was spread evenly across all treatment groups. Table 2 summarizes the health status across treatment groups. There were no significant differences in terms of health status between vaccination programs. There were only 3 mortalities reported during the trial of which only one was deemed to be caused by infectious agents. One animal was euthanized following the occurrence of a fractured leg and the other died of bloat.

Hemophilus somnus Titre Results

Figure 1 summarizes the titre responses of each treatment group at each sampling period.

Within Treatment Comparisons:

Treatment A calves were vaccinated at branding with Somnu-Star Ph and vaccinated at weaning with Somnu-Star Ph combined with Starvac 4 Plus. *H. somnus* titres in these calves were significantly increased at weaning and significantly increased following a booster vaccination at weaning.

Treatment B calves were vaccinated at branding with Starvac 4 Plus and vaccinated at weaning with with Starvac 4 Plus. There was no significant change in *H. somnus* titre at any of the sample times

Treatment C calves were vaccinated at branding with Somnu-Star Ph combined with Starvac 4 Plus and vaccinated at weaning with Somnu-Star Ph combined with Starvac 4 Plus. *H. somnus* titres were not significantly increased at weaning but post-weaning titre was significantly higher than the branding titre.

Treatment D calves received no spring vaccination, and were vaccinated at weaning with Somnu-Star Ph combined with Starvac 4 Plus, a booster vaccination with Somnu-Star combined with Starvac BRSV was administered 3 weeks later. There was no significant difference in *H. somnus* titre at the first 3 sampling times, *H. somnus* titre increased significantly following the post-weaning booster.

Between Treatment Comparisons:

Branding Titre, There was no significant difference in titre between the treatment groups.

Weaning Titre, There was no significant difference in titre between the treatment groups.

3 Week Post Weaning Titre, Treatment A calves had a significantly higher *H. somnus* titre than Treatment B calves. Treatment C and D calves were not significantly different than either of the other groups.

Mannheimia hemolytica LeukotoxinTitre Results:

Figure 2 summarizes the Mannheimia hemolytica leukotoxin titre results.

Within Treatment Comparisons:

Leukotoxin titres increased for all treatments at each bleed except for Treatment D calves, where there was no significant increase in leukotoxin titre between the spring bleed and weaning, the titre did increase significantly following vaccination at weaning. The titre was higher at weaning than in the spring and increased significantly again following the booster at weaning. The leukotoxin titre increased significantly following weaning in calves that were not vaccinated as well.

Between Treatment Comparisons:

There were no differences in leukotoxin titres between treatments in the spring or at weaning. Following the booster vaccination at weaning Treatment A calves had significantly higher leukotoxin titres than calves that had not received Somnu-Star Ph Treatment B, the other 2 treatment groups were not significantly different from Treatment A or B.

Modified Live vs Killed Vaccination Results:

Tables 3, 4, 5 and 6 summarize the results of the IBR titres, BRSV titres, BVD type 1 and 2 titres. There were significant differences in post vaccination titres between treatment groups in all instances. Calves receiving the killed vaccine after having received modified live vaccines at weaning consistently had significantly higher titre responses to all 4 antigens.

Discussion:

Pre-vaccination calf programs have been promoted by a number of pharmaceutical companies in the past few years. These programs usually rely on a spring calfhood vaccination which is meant to "prime" the immune system of the calf so that vaccinations at weaning in the feedlot would have a better and faster response due to immunological memory. Another potential benefit might be to mitigate the occurrence of enzootic pneumonia or "summer pneumonia" of young calves on pasture as their immunity wains. However, there are relatively few field trials that have documented this immune response and whether or not it is protective.

In this group of calves, there were relatively few treatments for bovine respiratory disease. These treatments were relatively even across the various vaccination programs and no specific differences were detected. As well, there were no differences in weaning weight performance between the vaccination programs. There were virtually no cases of enzootic or summer pneumonia, and so it was impossible to compare this across treatment groups. There did not appear to be any significant differences in respiratory disease treatments across the various vaccination programs although with so few treated, any real differences would have been hard to detect in a trial of this size.

There were no significant differences in serological titres between the various vaccination programs at either branding or weaning. However, Treatment A was consistently higher in terms of titres to Hemophilus somnus and Mannheimia hemolytica leukotoxin at post weaning. These calves received Somnustar PH at branding time in the spring and received it again combined with Starvac 4 Plus at weaning time. This would suggest that the calves that received this vaccine in the spring successfully primed their immune system and then responded with a higher secondary immune response when revaccinated in the fall. Treatment C which received Somnustar PH with StarVac 4 Plus in both the spring and the fall, however, was not statistically significantly higher than the control groups although their titres tended to be slightly higher as well.

When compared with the final titres of Treatment D (calves which received no spring vaccinations but received two vaccinations of Somnustar PH at weaning and 3 weeks later), it would appear that the immune response of the Treatment A calves is similar in strength to calves that receive the Somnustar PH at weaning and administered a booster vaccination 3 weeks later.

Most feedlots are reluctant to run cattle 3 weeks after arrival to receive a booster vaccination and tend to rely on one vaccination at arrival only. If this difference in titers is significantly more protective, this would suggest that feedlots would benefit from buying calves that have already received one dose of a Hemophilus somnus/Mannheimia hemolytica vaccine prior to arrival and then boostering them with the similar vaccine once they arrive. This also has major implications in terms of disease prevention as the vast majority of bovine respiratory disease treatments occur during the first 3 weeks of the feeding period. If protective titers can be derived earlier in the feeding period by priming the calf's immune system with a calfhood vaccination, this could have significant effects on feedlot morbidity and mortality.

Calves vaccinated with Starvac 4Plus, a MLV vaccine, at spring turnout and again at weaning maintained antibody levels in the low to moderate range for at least 5 months. BVD titre on entry to the feedlot is correlated with a decrease in "pulls" for Bovine Respiratory Disease. The results also confirm that a killed viral vaccine can give a constantly greater antibody response in calves that have been previously vaccinated with a modified live vaccine and in the face of high circulating antibody level.

Antibody response is only part of the immune response, measuring the cell-mediated response is relatively complex when compared to measuring antibody response. Recent publications have shown calves are capable of making a cell-mediated response in the presence of high circulating antibody without a detectable antibody response. Vaccinating with a MLV in the face of high circulating antibody may induce a cell-mediated response, which was not measured in this trial.

Vaccinating backgrounded calves that have already been vaccinated with modified live vaccines with an oil adjuvanted inactivated viral vaccine when they enter the feedlot appears to be the protocol of choice. Vaccinating replacement

heifers prior to breeding with an inactivated vaccine has also been shown to initiate an effective antibody response. Using an inactivated vaccine in pre-breeding protocols when an effective MLV program has been employed as calves has an advantage as there is no effect on the reproductive organs and vaccination can be done closer to the start of breeding season.

The most effective vaccination protocol appears to be a combination of MLV and inactivated vaccines.

Vaccines:

Starvac 4Plus Novartis Animal Health Canada Inc.
Plaza 3 – 2000 Argentia Road
Mississauga, Ontario L5N 1V9

Vira Shield 5 Novartis Animal Health Canada Inc.
Plaza 3 – 2000 Argentia Road
Mississauga, Ontario L5N 1V9

Project Activities

A scientific paper on the results of this trial is currently being prepared for publication. More serological titer results on IBR, BVD type 1 and 2, and BRSV are pending. Dr. Keith West of Prairie Diagnostic Services was involved in an unfortunate car accident and there have been some delays in the delivery of additional titer results. Plans are to submit the results to the Canadian Veterinary Journal and to present results to the Western Canadian Association of Bovine Practitioners. Presentations on the results to producer groups are planned for several venues including the Western Beef Development Cow-Calf School. The results of this trial have resulted in Novartis Animal Health funding another project which examines the combination of live and killed vaccines in feedlot cattle which is currently in progress.

Information of benefit to producers

This trial demonstrates the benefits of utilizing a Mannheimia hemolytica leukotoxin/Hemophilus somnus vaccine in a calfhood vaccination program. Although there was no enzootic pneumonia present in this group to evaluate the effects on young calves, it would appear that giving Somnustar PH (Novartis Animal Health Canada Inc.) does prime the immune system so that these calves respond with memory to a second vaccination at weaning. This has implications in terms of providing value to the feedlot in terms of prevaccination programs and also has a benefit to producers who are retaining ownership of calves.

It also demonstrated that in calves who have already received a modified live viral vaccine that a booster vaccination with a killed vaccine produces a significantly higher titer response than a booster vaccination with a modified live

vaccine. This has significant implications for feedlots who are purchasing pre-vaccinated calves. It also has an implication for heifer replacement vaccination programs and perhaps even adult cow vaccination programs. These findings need to be investigated in greater detail.

Table 1: Vaccinations given to calves in each treatment group

TIMING	Spring Branding		Fall Weaning		Post-Weaning	
Date	June 12 & 13		Oct 1 & 2		Oct 23 & 24	
VACCINE	SS-PH	SV4	SS-PH	SV4	SS-PH	SV4
Treatment A	X		X	X		
Treatment B		X		X		
Treatment C	X	X	X	X		
Treatment D			X	X		X

SS-PH: (Somnustar-PH, Novartis) A killed vaccine which contains antigens for Hemophilus somnus and Pasteurella hemolytica

SV4: (StarVac 4 Plus, Novartis) A modified live vaccine which contains antigens for bovine viral diarrhea virus type I and II, Infectious bovine rhinotracheitis virus, bovine respiratory syncytial virus and parainfluenza 3 virus.

Table 2: Morbidity results by Treatment Group

	Treatment A	Treatment B	Treatment C	Treatment D
Spring Vaccine	SS-PH	SV4	SS-PH + SV4	None
Fall Vaccine	SS-PH+ SV4	SV4	SS-PH + SV4	SSPH + SV4
Number	72	75	73	71
BRD Treatments	7 (9.7%)	8 (10.6%)	10 (13.7%)	3 (4.3%)
Foot Rot Treatments	4	0	6	4
Other Treatments	1	1	1	3

SS-PH: (Somnustar-PH, Novartis) A killed vaccine which contains antigens for Hemophilus somnus and Pasteurella hemolytica

SV4: (StarVac 4 Plus, Novartis) A modified live vaccine which contains antigens for bovine viral diarrhea virus type I and II, Infectious bovine rhinotracheitis virus, bovine respiratory syncytial virus and parainfluenza 3 virus.

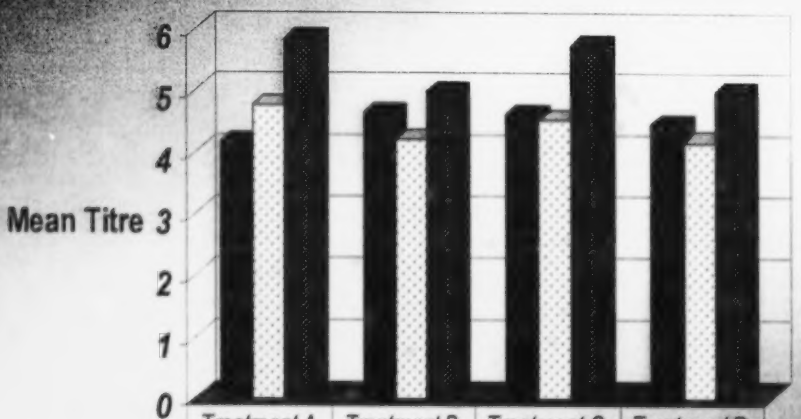
Table: 3	IBR ELISA Titres			
	Control	MLV	**Killed**	P
N	5	20	20	
Pre-Vaccination	22.2	35.7	24.6	P=0.3002
Post-Vaccination	21.4[a]	71.0[a]	103.2[b]	P=0.0000
Titre Change	-0.83[a]	35.3[b]	78.6[c]	P=0.0000

Table: 4	BRSV ELISA Titres			
	Control	**MLV**	**Killed**	**P**
N	5	20	20	
Pre-Vaccination	44.9	46.5	37.5	P=0.2312
Post-Vaccination	43.6[a]	60.3[a]	121.6[b]	P=0.0000
Titre Change	-5.8[a]	13.8[b]	84.10[c]	P=0.0000

Table: 5	Geometric Mean Titre BVD Type 1			
	Control	**MLV**	**Killed**	**P**
N	5	20	20	
Pre-Vaccination	280[a]	1,595[b]	1,822[b]	P < 0.05
Post-Vaccination	183[a]	2,223[a]	11,958[b]	P=0.0000
Titre Change	97[a]	628[a]	10,136[b]	P=0.0000

Table: 6	Geometric Mean Titre BVD Type 2			
	Control	**MLV**	**Killed**	**P**
N	5	20	20	
Pre-Vaccination	65.7[a]	266.3[a]	1,531[b]	P=0.0007
Post-Vaccination	55.2[a]	1589[a]	12,313[b]	P=0.0000
Titre Change	-10.5[a]	1,322.7[b]	10,782[b]	P=0.0000

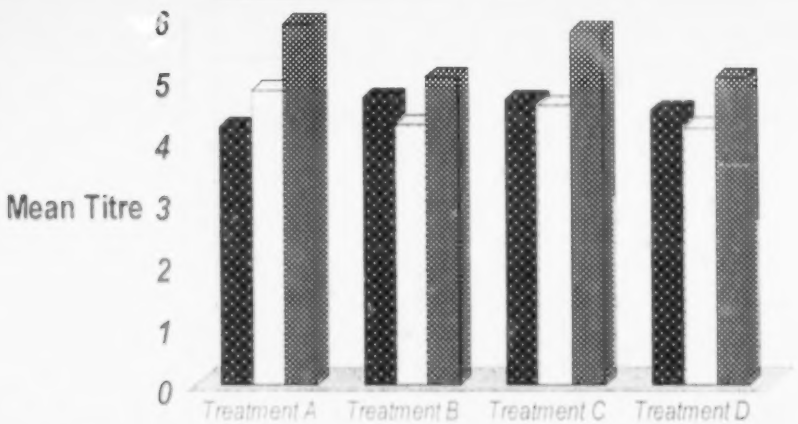
Figure 1: Hemophilus somnus mean titres



	Treatment A	Treatment B	Treatment C	Treatment D
Spring	4.1668	4.6412	4.6162	4.444
Weaning	4.7629	4.2209	4.5382	4.1749
Post-weaning	5.8393	4.9824	5.7302	4.9774

Treatment groups

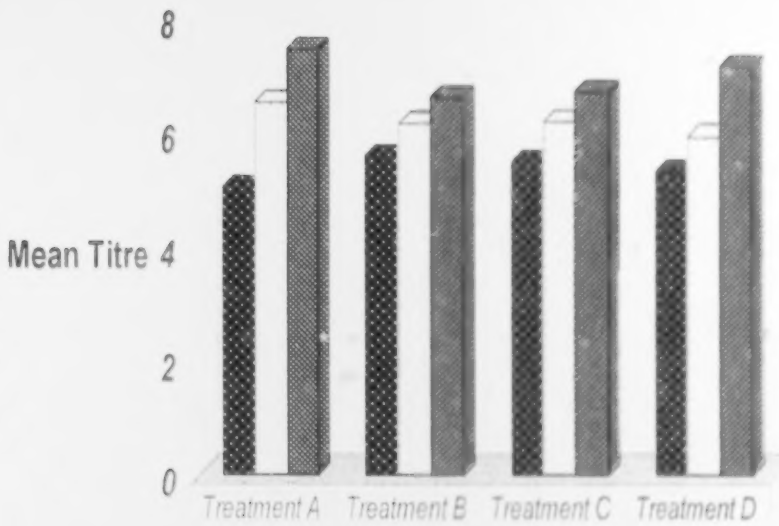
Figure 1: Hemophilus somnus mean titres



	Treatment A	Treatment B	Treatment C	Treatment D
Spring	4.1666	4.6412	4.6162	4.444
Weaning	4.7629	4.2209	4.5382	4.1749
Post-weaning	5.8393	4.9824	5.7302	4.9774

Treatment groups

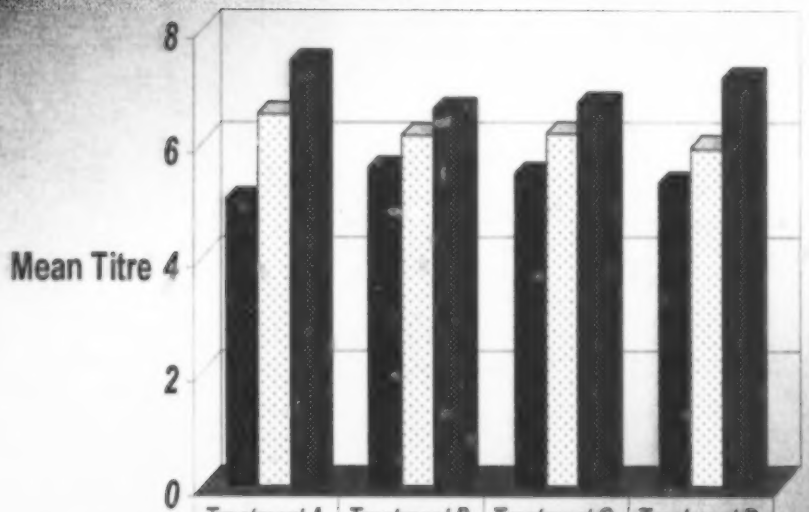
Figure 2:Mannheimia hemolytica Leukotoxin Mean Titres



	Treatment A	Treatment B	Treatment C	Treatment D
Spring	5.0268	5.5581	5.4602	5.294
Weaning	6.4996	6.1272	6.158	5.9161
Post-weaning	7.4284	6.5867	6.6914	7.1144

Treatment Groups

Figure 2:Mannheimia hemolytica Leukotoxin Mean Titres



	Treatment A	Treatment B	Treatment C	Treatment D
Spring	5.0268	5.5581	5.4602	5.294
Weaning	6.4996	6.1272	6.158	5.9161
Post-weaning	7.4284	6.5867	6.6914	7.1144

Treatment Groups